

What is claimed is:

1           1.       A method of fabricating a microfluidic device, comprising:  
2           providing first and second substrate layers;  
3           fabricating a microscale groove into at least a first surface of at least one of the first  
4 and second layers, and concurrently fabricating an alignment structure into the at least one surface  
5 of the first or second layers at a desired position relative to the microscale groove;  
6           mating one or more of a third component of the microfluidic device and a tool with  
7 the alignment structure to align the third component or the tool relative to the microscale groove.

1           2.       The method of claim 1, wherein the first substrate comprises a silica-based  
2 substrate and the alignment structure is etched into the first surface of the first substrate.

1           3.       The method of claim 1, wherein the first substrate comprises a polymeric  
2 substrate and the alignment structure is embossed onto the first surface of the first substrate.

1           4.       The method of claim 1, wherein the first substrate comprises a polymeric  
2 substrate and the alignment structure is injection molded onto the first surface of the first substrate.

1           5.       The method of claim 1, wherein the alignment structure comprises a  
2 depression on the first surface, and the tool comprises a drill.

1           6.       The method of claim 1, wherein the third component comprises a capillary,  
2 and the alignment structure comprises an aperture or well that is configured to receive the capillary  
3 element.

1           7.       The method of claim 25, wherein the alignment structure comprises a notch  
2 at an edge of the first surface of the first substrate, the first groove terminating in the notch, the  
3 notch being sized to receive the capillary element and such that a capillary channel disposed  
4 through the capillary element is in fluid communication with the groove.

1                   8.       The method of claim 26, wherein the second substrate layer comprises a  
2 second notch fabricated into a surface thereof at an edge of the surface of the second substrate layer,  
3 the second notch being positioned to correspond with the notch in the first substrate when the first  
4 and second substrates are mated together.

1                   9.       The method of claim 26, wherein the capillary element is inserted into an  
2 aperture created by the notch in the first substrate and the notch in the second substrate when the  
3 first and second substrates are mated, the capillary element operating as an alignment key during a  
4 step of bonding the first and second substrates together.

1                   10.     A method of fabricating a multilayered microfluidic device, comprising:  
2 providing a first notch in a first substrate layer;  
3 providing a second notch in a second substrate layer, the first and second notches  
4 being positioned to be complementary when the first and second substrate layers are mated  
5 together;  
6 inserting an alignment key into one of the first and second notches, the alignment  
7 key being configured to fit into the first and second notches when the first and second substrate  
8 layers are mated together, and align the first and second substrate layers in a first relative position;  
9 and  
10 mating and bonding the first substrate layer to the second substrate layer in the first  
11 relative position.

1                   11.     The method of claim 10, wherein the first and second notches are disposed at  
2 the edges of the first and second substrate layers, respectively.

1                   12.     The method of claim 11, wherein the one of the first and second substrates  
2 comprises a groove fabricated into a surface thereof, the groove terminating in one of the first or  
3 second notches, and wherein the alignment key comprises a capillary element.  
4

1                   13.     The method of claim 12, wherein the capillary element comprises a  
2 rectangular capillary having a capillary channel disposed therethrough.